Remarks

Reconsideration of this Application is respectfully requested.

At page 3, paragraph 3 of the Office Action, the Examiner requests an election of one invention to prosecute in the above-mentioned application. The Applicants hereby elect to prosecute the invention of Group I, represented by claims 1-13 and 18-23. This election is made without prejudice or disclaimer of the other claims or invention disclosed. This election is made without traverse.

Upon entry of the foregoing amendment, claims 1, 6-8 and 18-21 are pending in the application, with 1 and 18 being the independent claims. Claims 2-5, 9-17, 22 and 23 are sought to be cancelled without prejudice to or disclaimer of the subject matter therein. These changes are believed to introduce no new matter, and their entry is respectfully requested.

Based on the above amendment and the following remarks, Applicants respectfully request that the Examiner reconsider all outstanding objections and rejections and that they be withdrawn.

Claim Objections

At page 4 of the Office Action, the Examiner has objected to claims 7, 8, 12, 20, and 21 because of the recitation of "*pgi.*" Applicants have amended claim 7 to define "*pgi*" as "phosphoglucose isomerase," thereby clarifying the recitation of "*pgi*" in claims 8, 20 and 21. Claim 12 has been cancelled. Therefore, withdrawal of this objection is respectfully requested.

Rejections under 35 U.S.C. § 112, Second Paragraph

At page 4 of the Office Action, the Examiner has rejected claim 2-5, 9, 10 and 12 under 35 U.S.C. § 112, second paragraph, as allegedly being indefinite for failing to particularly point out and distinctly claim the subject matter which applicants regard as the invention. Applicants have cancelled these claims. Accordingly, withdrawal of this rejection is respectfully requested.

At page 5, paragraph 1 of the Office Action, the Examiner alleges that the term in claim 12 "a gene selected from the group consisting of a mutated *pgi* gene" is unclear and confusing. Applicants have cancelled this claim. Therefore, withdrawal of this rejection is respectfully requested.

Rejections under 35 U.S.C. § 112, First Paragraph

At page 5 of the Office Action, the Examiner has rejected claims 1-13 and 18-23 under 35 U.S.C. § 112, first paragraph, as allegedly containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. Applicants respectfully traverse this rejection.

Applicants have amended claims 1, 6-8 and 18-21 such that the scope of the claims is now drawn to a method of producing amino acids selected from the group consisting of L-lysine, L-threonine, and L-isoleucine comprising: culturing a *Corynebacterium glutamicum* cell with a mutant *pgi* gene. Amended claims 1, 6-8, and 18-21 are described in full, concise and exact terms that a skilled artisan would recognize that Applicants were in possession of the claimed invention. Support for these claims can be found in the

following sections of the specification: page 4, lines 3 and 23; page 6, lines 24 and 28; page 9, line 3; page 10, line 13; page 11; line 9; and pages 29-30. Applicants have cancelled claims 2-5, 9-13, 22, and 23. Therefore, withdrawal of this rejection is respectfully requested.

At page 6, paragraph 2 of the Office Action, the Examiner has rejected claims 1-7, 9-13, and 18-20 under 35 U.S.C. §112, first paragraph as allegedly failing to enable any person skilled in the art to which the invention pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims. Applicants respectfully traverse this rejection.

Specifically, the Examiner states that the specification

> does not reasonably provide enablement for a method of producing L-amino acids comprising: culturing an altered bacterial cell with <u>any</u> *pgi* gene mutation or other alterations producing an increased level of NADPH compared to an unaltered cell, wherein L-amino acid yields from the altered bacterial cell are greater than yields from an unaltered cell.

Office Action at page 7.

Applicants have limited the scope of claims 1, 6, 7, and 18-20 to a method of producing L-lysine, L-threonine and/or L-isoleucine comprising: culturing an altered *Corynebacterium glutamicum* cell with a mutant *pgi* gene. This method is fully described by the specification as filed at page 10, line 3; and pages 29-30. Claims 2-5 and 9-13 have been cancelled. Therefore, withdrawal of this rejection is respectfully requested.

Rejections under 35 U.S.C. § 102, Second Paragraph

At page 9, paragraph 2 of the Office Action, the Examiner has rejected claims 1, 2, 4-7 and 18-20 under 35 U.S.C. §102(b) as anticipated, or in the alternative, as obvious under 35 U.S.C. §103(a). Please note that the obviousness rejection is discussed below.

With respect to the §102(b) rejection, the Examiner alleges that claims 18-20 are anticipated by Mascarenhas *et al.* Specifically, Mascarenhas *et al.* teach a method of producing L-tryptophan by using a genetically altered strain of *Escherichia coli* with a deleted *pgi* gene. They show that the deletion of the *pgi* gene increased the average rate of tryptophan production by a 2:1 ratio.

Mascarenhas *et al.* is strictly limited to a method of producing aromatic amino acids and does not teach a method of producing L-lysine, L-threonine and/or L-isoleucine as is now claimed. Furthermore, the method in Mascarenhas *et al.* teaches the use of *Escherichia coli*, whereas the present invention is limited to the use of *Corynebacterium glutamicum.* Accordingly, the method used in Mascarenhas *et al.* does not contain each element as set forth in claims 18-20 as written. The withdrawal of this rejection is respectfully requested.

Rejections under 35 U.S.C. § 103

At page 9, paragraph 2, the Examiner alleges that claims 1, 2, and 4-7 are rejected under 35 U.S.C. § 103(a) as obvious over Mascarenhas *et al.* Applicants respectfully traverse this rejection. Specifically, the Examiner states that

> Mascarenhas *et al.* suggest that deleting the *pgi* gene alters the path of glucose catabolism diverting carbon through the...pentose phosphate pathway...[the result of which is] the accelerated generation of NADPH, which serves to drive the biosynthesis of aromatic intermediates and glutamate.

Office Action at page 10.

Mascarenhas *et al.* suggest only that the deletion of the *pgi* gene may have an effect on the biosynthetic capabilities of *Escherichia coli* for the production of aromatic intermediates. Office Action at page 10. Applicants have amended the scope of claims 1,

6 and 7 to encompass a method of producing an amino acid selected from the group consisting of L-lysine, L-threonine and L-isoleucine comprising: culturing an altered *Corynebacterium glutamicum* cell having an increased amount of NADPH as compared to an unaltered *Corynebacterium glutamicum* cell, wherein yields of an amino acid selected from the group consisting of L-lysine, L-threonine and L-isoleucine from said altered *Corynebacterium glutamicum* cell are greater than yields from an unaltered *Corynebacterium glutamicum* cell. The fact that one particular bacterial strain is capable of producing aromatic intermediates is not an indication that a completely different organism will produce L-lysine, L-threonine and/or L-isoleucine. Based on the language in Mascarenhas *et al.*, an ordinary person skilled in the art would not have had a reasonable expectation of success of producing L-lysine, L-threonine and/or L-isoleucine using *Corynebacterium glutamicum* as claimed. Therefore, at the time the invention was made, it would not have been obvious to an ordinary person skilled in the art to use *Corynebacterium glutamicum* to produce L-lysine, L-threonine and/or L-isoleucine as claimed. Claims 2, 4 and 5 have been cancelled. Accordingly, withdrawal of this rejection is respectfully requested.

At page 11, paragraph 1 of the Office Action, the Examiner alleges that claims 3, 9-13, 22, and 23 are rejected under 35 U.S.C. § 103(a) as being obvious over Mascarenhas *et al.*, in view of Ischino *et al.*, Marx *et al.*, and Voet *et al.* Applicants have cancelled these claims and respectfully request the withdrawal of this rejection.

At page 14, paragraph 3 of the Office Action, the Examiner alleges that claims 8 and 21 are rejected under 35 U.S.C. §103(a) as being unpatentable over Mascarenhas *et al.* in view of Fitzpatrick *et al.* Obviousness cannot be established absent some teaching,

suggestion or incentive, and thus, even if it was obvious to one skilled in art to try various combinations to achieve the claimed methods (and Applicants do not believe it was), such evidence does not establish a *prima facie* case of obviousness. See *In re Geiger*, 2 USPQ2d 1276 (Fed. Cir. 1987) and *In re Fine*, 5 USPQ2d 1596 (Fed. Cir. 1988). Furthermore, in order to establish obviousness, the Examiner must show that the prior art suggested the modification of the reference or references required to arrive at the claimed invention, and that the invention could be attained with a reasonable expectation of success. See *In re Vaeck*, 20 U.S.P.Q.2d 1438, 1442 (Fed. Cir. 1992).

As previously mentioned, Mascarenhas *et al.* teach the production of aromatic intermediates using *pgi*-deficient *Escherichia coli*. The present invention is directed to a method of producing L-lysine, L-threonine and/or L-isoleucine using *Corynebacterium glutamicum*, which is not disclosed in Mascarenhas *et al.* Fitzpatrick *et al.* generally teach only a method of subcloning a *recA* gene into the genome of *Corynebacterium glutamicum* for the purpose of gene silencing and, therefore, does not cure the deficiencies of Mascarenhas *et al.* Fitzpatrick *et al.* is silent as to increasing the amounts of NADPH in *Corynebacterium glutamicum* to increase amino acid yields. Specifically, Fitzpatrick *et al.* does not teach or even suggest culturing *Corynebacterium glutamicum* cells with increased amounts of NADPH to produce the claimed amino acids. Therefore, the combination of these references does not render the invention obvious.

Furthermore, Fitzpatrick *et al.* teach away from subcloning genes into *Corynebacterium glutamicum*. In Fitzpatrick *et al.*, the authors note that "[n]atural recombination systems have hampered some efforts to construct stable vectors in coryneform bacteria by causing plasmid rearrangements and deletions" because coryneform

bacteria have high recombinatorial activity and genetic instability. (Fitzpatrick p. 575). A prior art reference that "teaches away" from the claimed invention is a significant factor to be considered in determining obviousness. *In re Gurley*, 31 USPQ2d 1130, 1132 (Fed. Cir. 1994); *see In re Geisler*, 43 USPQ2d 1362, 1366 (Fed. Cir. 1997). Thus, subcloning an internal region of the *pgi* gene and inserting the resulting vector into a *Corynebacterium glutamicum* is not obvious since the inventors would have had no reasonable chance of success based upon the language in Fitzpatrick *et al.*

For the foregoing reasons, the invention is not obvious in view of Mascarenhas *et al.*, and the addition of Fitzpatrick *et al.* does not add anything to render the invention unpatentable. Therefore, withdrawal of this rejection is respectfully requested.

Conclusion

All of the stated grounds of objection and rejection have been properly traversed, accommodated, or rendered moot. Applicants therefore respectfully request that the Examiner reconsider all presently outstanding objections and rejections and that they be withdrawn. Applicants believe that a full and complete reply has been made to the outstanding Office Action and, as such, the present application is in condition for allowance. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Prompt and favorable consideration of this Amendment and Reply is respectfully requested.

Respectfully submitted,

STERNE, KESSLER, GOLDSTEIN & FOX P.L.L.C.

Cynthia M. Bouchez
Agent for Applicants
Registration No. 47,438

Date: August 6, 2001

1100 New York Avenue, N.W.
Suite 600
Washington, D.C. 20005-3934
(202) 371-2600

Version with markings to show changes made

1. (Once Amended) A method of producing [L-] an amino acid[s] selected from the group consisting of L-lysine, L-threonine and L-isoleucine comprising:

culturing an altered [bacterial] *Corynebacterium glutamicum* cell having an increased amount of NADPH as compared to an unaltered [bacterial] *Corynebacterium glutamicum* cell, wherein [L-amino acid] yields of an amino acid selected from the group consisting of L-lysine, L-threonine and L-isoleucine from said altered [bacterial] *Corynebacterium glutamicum* cell are greater than yields from an unaltered [bacterial] *Corynebacterium glutamicum* cell.

6. (Once Amended) The method of claim 1, wherein said L-amino acid yields from said altered [bacterial] *Corynebacterium glutamicum* cell are from about 1% to about 100% greater than from said unaltered [bacterial] *Corynebacterium glutamicum* cell.

7. (Once Amended) The method of claim 1, wherein said altered [bacterial] *Corynebacterium glutamicum* cell has a mutant phosphoglucose isomerase *(pgi)* gene.

8. (Once Amended) The method of claim 1, wherein said altered [bacterial] *Corynebacterium glutamicum* cell is produced by

(a) subcloning an internal region of a *pgi* gene; and

(b) inserting said resulting vector from step (a) into a [bacterial] *Corynebacterium glutamicum* genome via homologous recombination.

18. (Once Amended) A method of producing L-amino acids selected from the group consisting of L-lysine, L-threonine and L-isoleucine, comprising:

culturing an altered [bacterial] *Corynebacterium glutamicum* cell having a decreased amount of 6-phosphoglucose isomerase enzymatic activity as compared to an unaltered [bacterial] *Corynebacterium glutamicum* cell wherein L-amino acid yields from said altered [bacterial] *Corynebacterium glutamicum* cell are greater than yields from an unaltered [bacterial] *Corynebacterium glutamicum* cell.

19. (Once Amended) The method of claim 18, wherein said L-amino acid yields from said altered [bacterial] *Corynebacterium glutamicum* cell are from about 1% to about 100% greater than from said unaltered [bacterial] *Corynebacterium glutamicum* cell.

20. (Once Amended) The method of claim 18, wherein said altered [bacterial] *Corynebacterium glutamicum* cell has a mutant *pgi* gene.

21. (Once Amended) The method of claim 18, wherein said altered [bacterial] *Corynebacterium glutamicum* cell is produced by

(a) subcloning an internal region of a *pgi* gene; and

(b) inserting said resulting vector from step (a) into a [bacterial] *Corynebacterium glutamicum* genome via homologous recombination.